AMYLOVORAN

WILLIAM F. FETT
Eastern Regional Research Center
Philadelphia, Pennsylvania

Amylovoran (originally referred to as amylovorin) is an acidic, high molecular mass (>1 MDa) bacterial extracellular polysaccharide (EPS) (1). It is produced by the plant pathogen *Erwinia amylovora*, the causal agent of fire blight on pome fruit trees and other rosaceous plants. On its rosaceous plant hosts, this necrogenic pathogen invades cortical tissues resulting in canker formation, colonizes the xylem leading to wilting of young shoots, and eventually causes the characteristic blackening of severely infected twigs, flowers, and foliage (2). Amylovoran can be present both as a tightly held capsule and a loosely held slime. It is composed of a branched pentasaccharide repeating unit consisting of D-galactosyl and D-glucuronyl acid residues in a molar ratio of 4:1. The terminal galactose moiety on the side branch is substituted with pyruvate and variable amounts of 2-linked, 3-linked and 2,3-linked acetate (Fig. 1) (3). Approximately 10% of the core α-D-galactosyl residues are linked to an additional D-glucosyl residue. Amylovoran synthesis is favored by the presence of abundant, easily utilizable carbohydrates such as sorbitol and galactose (4). Amylovoran synthesis is also stimulated by environmental stress such as the presence of copper sulfate (2 mm) in the medium (5) and temperatures less than optimal for growth. When sucrose is abundant, *E. amylovora* produces a second EPS, the fructose homopolymer levan.

The molecular genetics of amylovoran synthesis is the subject of intensive study (6). Amylovoran biosynthesis is encoded by the *ams*-region, where 12 genes (*amsA* to *amsL*) are transcribed as an operon producing a 16-kb mRNA (7). The *ams* cluster is very similar to the *cps* cluster of *Pantoea stewartii* subsp. *stewartii* that encodes for the synthesis of the EPS stewartan (8). The regulation of amylovoran production by *E. amylovora* is similar to the regulation of the synthesis of the capsular EPS colanic acid produced by *Escherichia coli* as well as stewartan (9). These bacteria have in common a family of positive and negative regulators. EPS synthesis is controlled by at least

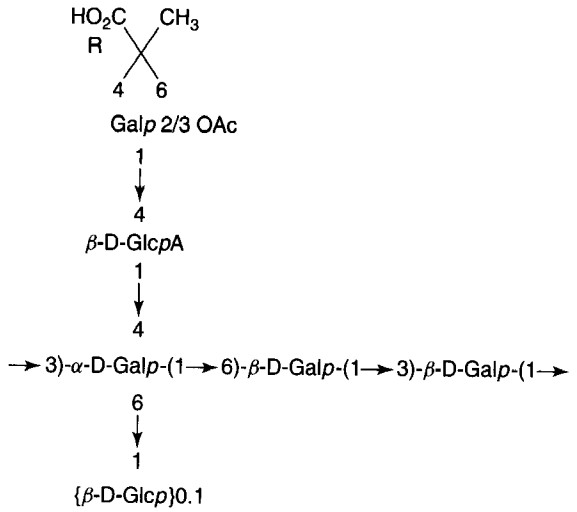

Figure 1. Structure of the repeating unit of amylovoran. Gal, galactose; GlcA, glucuronic acid; Glc, glucose (all three sugars are present in the pyranose form). The terminal galactosyl residue on the branch is either not *O*-acetylated (10%), mono-*O*-acetylated at the 2 (26%) or 3 (24%) positions, or di-*O*-acetylated at the 2 and 3 positions (40%). Approximately 10% of the α-D-galactosyl residues are linked to β-D-glucose (4).

two activator proteins, RcsA and RcsB, where RscB is part of a two-component regulatory system.

Several experimental observations indicate that amylovoran is required for full virulence of *E. amylovora*. First, amylovoran is produced in infected host plants. Bacterial ooze emanating from severely infected shoots and from the surfaces of inoculated pear slices is primarily composed of amylovoran, and bacteria present in the ooze have well-defined capsules (10). Ultrastructural studies indicate that amylovoran is produced in colonized xylem vessels (11). Second, treatment of shoots with amylovoran leads to non–host-specific wilting due to physical blockage of the xylem (12). Third, spontaneous uncharacterized transposon mutants of *E. amylovora* that appear nonmucoid on semisolid media are of reduced virulence (10). Finally, genetically defined amylovoran-deficient mutants either are of reduced virulence or are completely nonpathogenic (4,9,13).

BIBLIOGRAPHY

1. K. Jumel, K. Geider, and S.E. Harding, *Int. J. Biol. Macromol.* **4**, 251–258 (1997).
2. T. Van Der Zwet and H.L. Keil, *Fire Blight: A Bacterial Disease of Rosaceous Plants*, U.S. Department of Agriculture, Agriculture Handbook No. 510, Government Printing Office, Washington, D.C., 1979.
3. M. Nimtz et al., *Carbohydr. Res.* **287**, 59–76 (1996).
4. R C. Hignett, *Physiol. Mol. Plant Pathol.* **32**, 387–394 (1988).
5. S. Bereswill, S. Jock, P. Bellemann, and K. Geider, *Plant Dis.* 158–164 (1998).
6. T.P. Denny, *Annu. Rev. Phytopathol.* **33**, 173–197 (1995).
7. O. Kelm, C. Kiecker, K. Geider, and F. Bernhard, *Mol. Gen. Genet.* **256**, 72–83 (1997).
8. F. Bernhard et al., *Microbiology* **142**, 1087–1096 (1996).
9. S. Bereswill and K. Geider, *J. Bacteriol.* **179**, 1354–1361 (1997).
10. R.A. Bennett and E. Billing, *J. Gen. Microbiol.* **116**, 341–349 (1980).
11. C.G. Suhayda and R.N. Goodman, *Phytopathology* **71**, 697–707 (1981).
12. S.V. Beer, T.M. Sjulin, and H.S. Aldwinckle, *Phytopathology* **73**, 1328–1333 (1983).
13. F. Bernhard, D.L. Coplin, and K. Geider, *Mol. Gen. Genet.* **239**, 158–168 (1993).
14. M. Menggad and J. Laurent, *Eur. J. Plant Pathol.* **104**, 313–322 (1998).